Hydrobiologia (2006) 568:489–492
DOI 10.1007/s10750-006-0115-2

Primary Research Paper

Hydroponic versus rooted growth of *Zostera marina* L. (Eelgrass)

Patrick D. Biber[1,2,*]
[1]*Institute of Marine Science, University of North Carolina at Chapel Hill, 3431 Arendell Street, Morehead City, NC 28557, USA*
[2]*Gulf Coast Research Laboratory, The University of Southern Mississippi, 703 East Beach Drive, Ocean Springs, MS 39564, USA*
*(*Author for correspondence: Tel.: +1-228-872-4260; Fax: +1-228-872-4204; E-mail: Patrick.biber@usm.edu)*

Received 2 December 2005; in revised form 27 February 2006; accepted 5 March 2006; published online 3 May 2006

Key words: Eelgrass, hydroponic, seedling, growth

Abstract

Seagrass fragments and seeds are important dispersal mechanisms by which individuals can be transported to new habitats. While dispersal distances of these free-floating stages have been recently investigated in some detail, almost nothing is known about how long fragments or seedlings may remain viable in the water-column. This study reports on the results of an experiment in which both mature and seedling life-history stages of the temperate seagrass, *Zostera marina* L. were successfully maintained hydroponically over a 1-month-period. It is suggested that a potential application of this hydroponic growth approach could be seedling culture for restoration activities.

Short research note

The survival of fragments or seedlings during floating stages is of obvious importance for the dispersal of many seagrass species, however, this aspect of their biology has been poorly studied to date. Fragmentation can be an effective propagation and dispersal mechanism in temperate *Zostera marina* L., tropical *Halodule wrightii* Ascherson and endangered *Halophila johnsonii* Eiseman (Hall, 2002; Harwell & Orth, 2002). Rafting of dislodged reproductive shoots (=fragments) of *Z. marina* is a commonly observed phenomenon in the late spring in Chesapeake Bay (Orth et al., 1994). Harwell and Orth (2002) reported that reproductive shoots of *Z. marina* had the potential to disperse up to 100 km from their source bed and indicated that this may take 2–3 weeks. Given the lack of information reported in the literature on survival duration of free-floating fragments or seedlings, a pilot experiment to determine whether or not hydroponic growth is feasible in eelgrass, *Z. marina*, was undertaken.

Two life-history stages of *Z. marina* were grown for one month under two different environments: (1) hydroponic growth and, (2) rooted in sandy sediments. The two stages used were seedlings aged about 4–6 weeks post-germination and mature plants that were at least one growing season or more in age. These stages were easily distinguishable in the field at the time of collection. Seedlings were one shoot with 2–4 leaves, while mature fragments were standardized to two shoots, one of them being an apical shoot.

Mature plants and seedlings were collected from field sites (34° 45′ N, 76° 35′ W) in February 2003 during the early growing season of this species in North Carolina, USA (Nelson, 1979). Approximately half of the collected plants were transplanted within 24 h into 2 plastic tubs filled with medium-grained beach sand that had been dried and sieved to remove large debris; the remaining half of the plants were placed inside two clear plastic food-containers (2 l) with many small holes drilled in the sides and bottom to ensure water exchange. Tubs and containers were placed

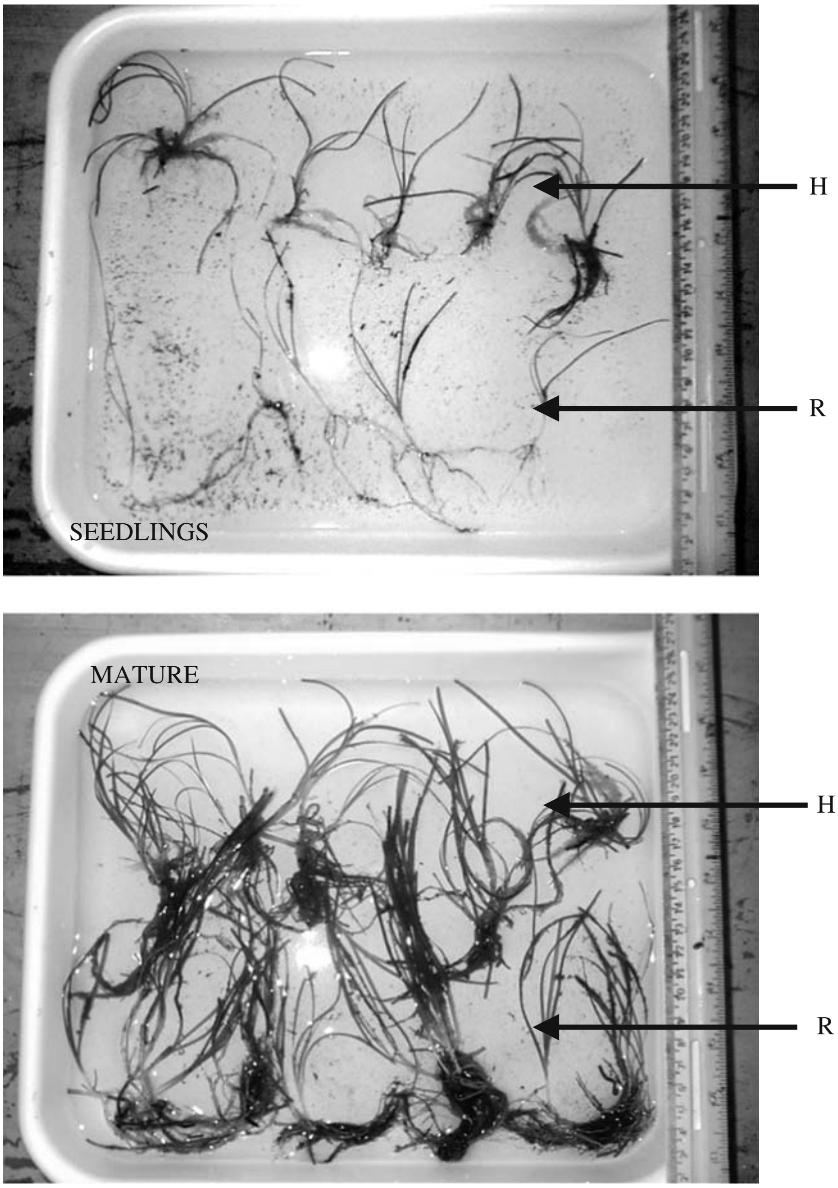

Figure 1. Photos of seedlings (top) and mature (bottom) plants after 1-month growth in hydroponic (H) chambers or growing rooted in sandy sediment (R) in the tank.

in a large shallow (depth = 0.5 m, volume = 5000 l) outdoor tank and maintained on flow-through filtered seawater that was pumped from a nearby tidal channel, with a turnover rate of about once per 3 days. The tank was shaded from direct sunlight with 50% shade cloth at a height of 2 m above the water (5.95 mol photons m^{-2} day^{-1}). In comparison to natural field conditions in the turbid estuarine waters of North Carolina, this approximates daily irradiances seen by plants that are growing at 1 m depth or less (7.5 mol photons m^{-2} day^{-1}).

After one month, random plants ($N = 5$ per treatment and stage) were removed from the two respective treatments. The number of shoots and leaves per plant were counted and the number of leaves per shoots calculated. The length of the longest leaf per plant (an approximation of canopy height) was measured from the base of the sheath to the tip of the leaf and I noted if the tip was intact or had been removed (caused by grazing, senescence, or physical damage), the width was also measured about half way along the length of the leaf. From these measures the leaf area (one-side) was calculated.

The second youngest leaf on the apical shoot (for seedlings this was the only shoot) was selected for photosystem II fluorescence yield measurements (Fv/Fm), as per the recommendations of Durako and Kunzelmann (2002). The central portion of the leaf was dark-adapted for 10 minutes using the clips provided with the OptiSciences OS-30 Plant Efficiency Analyser (PEA). When the leaves were ready, i.e. dark-adapted, the fluorescence over 4 s was recorded; from the base fluorescence (Fo) and the maximum fluorescence (Fm) the potential quantum yield ratio (Fv/Fm) was calculated using the method of Beer et al. (2001).

Hydroponic survival was observed to be a minimum of one month for two life-history stages in the temperate seagrass *Z. marina*, during the early growing season in North Carolina, when water temperatures were between 9 and 17 °C. Both seedlings and mature fragments were apparently healthy after 4 weeks floating in the water as evidenced by the increase in size and healthy photosynthetic yield results (Fv/Fm > 0.5).

Seedlings showed greater growth in the hydroponic treatment than the rooted treatment over the one month duration of the experiment (Fig. 1). Seedlings in the hydroponic treatment were found to have produced significantly more shoots ($p < 0.05$) and leaves ($p < 0.01$), and were also noted to have shorter (n.s.) and wider leaves ($p < 0.05$) than their counterparts rooted in the tubs (Tables 1 and 2). However, Fv/Fm ratios suggest that the seedlings in the hydroponic treatment had lower photosystem II performance, potentially indicative of reduced photosynthesis, although this was not significant for the sample size tested. In contrast, mature plants grown in the sandy sediment tended to be marginally larger, but not significantly so for any of the variables measured. Mature plants rooted in the sediment produced more shoots and leaves, and were also noted to have longer and narrower leaves compared to the hydroponic containers (Table 1). Fv/Fm ratios in the mature rooted plants were slightly less than the hydroponic plants. Mature plants in both treatments had higher photosystem II performance than seedlings (Table 1).

The hydroponic seedlings of *Z. marina* were noticeably larger after one month than their siblings growing in the sediment tubs, even though all other environmental conditions remained the same (Fig. 1). One possible explanation for these results is that early seedling growth involves the rapid development of roots, which possibly comes at the expense of leaf formation. In the hydroponic treatment, root growth cues may have been lacking allowing these plants to put more photosynthetic resources into leaf and shoot growth. A second factor that may have contributed to the larger sized seedlings is that hydroponic conditions may obviate sulfide stress that occurs in plants grown in anoxic sediments (Koch & Erskine, 2001). The hydroponic seedlings may have been able to grow to a larger size than those in the

Table 1. Mean (± SE), $N = 5$, of morphological variables and Fv/Fm measured on seedlings and mature plants of *Zostera marina* at the start (Initial) and after 1 month growth in two treatments (Rooted in tubs or Hydroponic), with the larger response underlined, * indicates significantly greater at alpha = 0.05

	No. shoots	No. leaves	Leaves/shoot	Length (mm)	Width (mm)	Area (cm^2)	Fv/Fm
Seedlings							
Initial	1.0 (0.00)	2.7 (0.21)	2.72 (0.211)	96.7 (8.21)	0.99 (0.037)	0.96 (0.100)	0.634 (0.013)
Rooted	1.0 (0.00)	2.8 (0.2)	2.80 (0.200)	<u>110.2 (11.73)</u>	1.18 (0.086)	1.31 (0.156)	<u>0.613 (0.012)</u>
Hydroponic	<u>1.6 (0.25)*</u>	<u>6.2 (0.86)*</u>	<u>4.00 (0.354)*</u>	97.2 (5.83)	<u>1.50 (0.084)*</u>	<u>1.45 (0.088)</u>	0.587 (0.018)
Mature							
Initial	2.8 (0.24)	11.0 (0.96)	4.05 (0.209)	131.1 (6.89)	1.93 (0.000)	2.53 (0.133)	0.666 (0.009)
Rooted	<u>4.4 (0.68)</u>	<u>17.2 (2.82)</u>	<u>3.89 (0.323)</u>	<u>137.0 (12.36)</u>	1.96 (0.136)	<u>2.74 (0.387)</u>	0.623 (0.005)
Hydroponic	3.2 (0.37)	11.2 (2.13)	3.42 (0.486)	125.0 (5.26)	<u>2.10 (0.084)</u>	2.63 (0.198)	<u>0.656 (0.027)</u>

Table 2. Results of *t*-tests (parametric) /Mann–Whitney *U*-tests (non-parametric) on rooted vs. hydroponic plants for the variables reported in Table 1

	No. shoots (n)	No. leaves (n)	Leaves/shoot (p)	Length, mm (p)	Width, mm (p)	Area, cm^2 (n)	Fv/Fm (p)
Seedlings	<u>0.0495</u>	<u>0.0071</u>	<u>0.0183</u>	0.35	<u>0.0285</u>	0.6752	0.267
Mature	0.1419	0.2101	0.4381	0.3978	0.4071	0.5296	0.2543

(n) indicates non-parametric test with *p* values from Chi-square distribution and (p) indicates a parametric *t*-test with *p* values from *t*-distribution. Underlined indicates significant result at alpha = 0.05

sediments because the photosynthetic energy needed to maintain an oxic rhizosphere in reducing sediments (Lee & Dunton, 2000) could instead be allocated to tissue growth (Koch et al., 1990).

Given the larger number of leaves and shoots observed in the hydroponic seedlings, this pilot study suggests that an interesting potential application could be alternative culture practices for nursery-raised seeds and seedlings, which could then be used in restoration activities. Clearly further research needs to be done to investigate the survival and growth of hydroponically cultured seedlings after planting; an important aspect of seagrass biology that was not investigated in this pilot study. The increased importance of seagrass restoration in areas where water-quality has returned to suitable conditions requires new ideas about propagation of seedlings to help improve on methods that have been attempted to date in the field of seagrass restoration (e.g., Bird et al., 1994; Fonseca et al., 1998).

References

Beer, S., M. Bjork, R. Gademan & P. Ralph, 2001. Measurements of photosynthetic rates in seagrasses. In Short, F. T., R. G. Coles & C. A. Short (eds), Global Seagrass Research Methods. Elsevier, Amsterdam, 183–198.

Bird, K. T., J. Jewett-Smith & M. S. Fonseca, 1994. Use of *in-vitro* propagated *Ruppia maritima* for seagrass meadow restoration. Journal of Coastal Research 10: 732–737.

Durako, M. J. & J. I. Kunzelman, 2002. Photosynthetic characteristics of *Thalassia testudinum* measured *in situ* by pulse-amplitude modulated (PAM) fluorometry: methodological and scale-based considerations. Aquatic Botany 73: 173–185.

Fonseca, M. S., W. J. Kenworthy & G. W. Thayer, 1998. Guidelines for the Conservation and Restoration of Seagrasses in the United States and Adjacent Waters. NOAA Coastal Ocean Program Decision Analysis Series No. 12. NOAA Coastal Ocean Office, Silver Spring, MD.

Hall, L. M., 2002. Fragments of the seagrasses *Halodule wrightii* and *Halophila johnsonii* as potential recruits in Indian River Lagoon, Florida. MS Thesis, Florida Institute of Technology, Melbourne, FL.

Harwell, M.C. & R.J. Orth, 2002. Long-distance dispersal potential in a marine macrophyte. Ecology 83: 3319–3330.

Koch, M. S., I. A. Mendelssohn & K. L. McKee, 1990. Mechanism for the hydrogen sulfide-induced growth limitation in wetland macrophytes. Limnology and Oceanography 35: 399–408.

Koch, M. S. & J. M. Erskine, 2001. Sulfide as a phytotoxin to the tropical seagrass *Thalassia testudinum*: interactions with light, salinity and temperature. Journal of Experimental Marine Biology and Ecology 266: 81–95.

Lee, K. S. & K. H. Dunton, 2000. Diurnal changes in pore water sulfide concentrations in the seagrass *Thalassia testudinum* beds: the effects of seagrasses on sulfide dynamics. Journal of Experimental Marine Biology and Ecology 255: 201–214.

Nelson, W. G., 1979. An analysis of structural pattern in an eelgrass (*Zostera marina* L.) amphipod community. Journal of Experimental Marine Biology and Ecology 39: 231–264.

Orth, R. J., M. Luckenback & K. A. Moore, 1994. Seed dispersal in a marine macrophyte: implications for colonization and restoration. Ecology 75: 1927–1939.